IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Appln No. : 10/600,266

Applicant(s): Fumitoshi ASAI et al.

Filed : June 20, 2003

For : MEDICINAL COMPOSITIONS CONTAINING ASPIRIN

Art Unit : 1614

Examiner : Brian Yong S. Kown

Docket No. : 03337C/HG

Confirm. No.: 7488

Customer No.: 01933

DECLARATION UNDER 37 CFR 1.132

Atsuhiro Sugidachi, declare that I am a co-inventor of the invention described and claimed in the above-referenced application.

1. I graduated from the Faculty of Pharmaceutical Sciences, Tohoku University, Japan, in 1987. I received a Ph.D. from the Faculty of Pharmaceutical Sciences of Tohoku University in 1996. I entered into the employment of Sankyo Co., Ltd., Tokyo, Japan, in April, 1989 and am now a senior chief researcher in Pharmacology and Molecular Biology Research Laboratories of the said company. From March, 2000 through February, 2002, I studied at the School of Medicine, the University of Pennsylvania, Philadelphia, USA.

I am a member of the following scientific societies:

The Japanese Pharmacological Society,

The Japanese Society of Thrombosis and Hemostasis.

Representatives of the scientific reports recently written by my co-workers and me are as recited below.

(1) "Antiplatelet action of R-99224, an active metabolite of a novel thienopyridine-type G_i-linked P2T antagonist, CS-747."; Br. J. Pharmacol. 132, 47-54 (2001)

(2) "Pharmacological profiles of R-96544, the active form of a novel 5-HT_{2A}, receptor antagonist R-102444."; Eur. J. Pharmacol. 457, 107-114 (2002)

(3) "Effects of R-102444, an orally active 5-HT_{2A} receptor antagonist, in rat models of peripheral vascular disease."; Vascular Pharmacology 41, 7-13 (2004)

(4) "Stereoselective inhibition of human platelet aggregation by R-138727, the active metabolite of CS-747 (Prasugrel, LY640315), a novel $P2Y_{12}$ receptor inhibitor."; Thromb. Haemost. 94, 593-598 (2005)

The following testing was accomplished under my supervision and control.

Comparison Testing

Ex vivo platelet aggregation was measured in platelet-rich plasma (PRP) from male Sprague-Dawley rats. 2-Acetoxy-5-(a-cyclopropylcarbonyl-2-fluorobenzyl)-4,5,6,7-tetrahydrothieno[3,2-

c]pyridine (hereinafter referred as Compound A, 3 and 6 mg/ml) and Aspirin (10 mg/ml) were suspended in a 5% (w/v) solution of gum arabic.

Each test compound in combination with or without aspirin was orally administered to non-fasted rats in a volume of 1 ml/kg. The rats were anesthetized with pentobarbital (40 mg/kg, *i.p.*) 0.5 hr after the oral dosing, and blood was collected using 3.8% (w/v) sodium citrate solution (1/9 of the blood, v/v) as an anticoagulant. The PRP was prepared by centrifuging the blood at 230xg for 15 min at room temperature. Platelet-poor plasma (PPP) was obtained by a subsequent centrifugation of the remaining blood at 2,000xg for 10 min at room temperature.

Platelet counts in PRP (platelet-rich plasma) were determined using a hematology analyzer and adjusted to $5x10^8$ platelets/ml by the addition of PPP. Platelet aggregation was measured with a platelet aggregometer. PRP (240 µl) was placed into a cuvette, and prewarmed for 1.5 min at 37°C, and then stimulated with 10 µl of collagen (final concentration of 20 µg/ml). Platelet aggregation was recorded for 7 min, and maximum aggregation was evaluated.

Results are expressed as the mean ± S.E.M. Statistical comparisons were carried out by Dunnett's test with respect to the control and by 2-way ANOVA for the assessment of any synergistic effect. A value of $p<0.05$ was considered to be statistically significant.